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A GPU-Accelerated Model of Neuroblastoma to Predict Disease Outcome and Find Drug Targets

Kenneth Y. Wertheim, Robert Chisholm, Paul Richmond, Dawn Walker

k.wertheim@sheffield.ac.uk

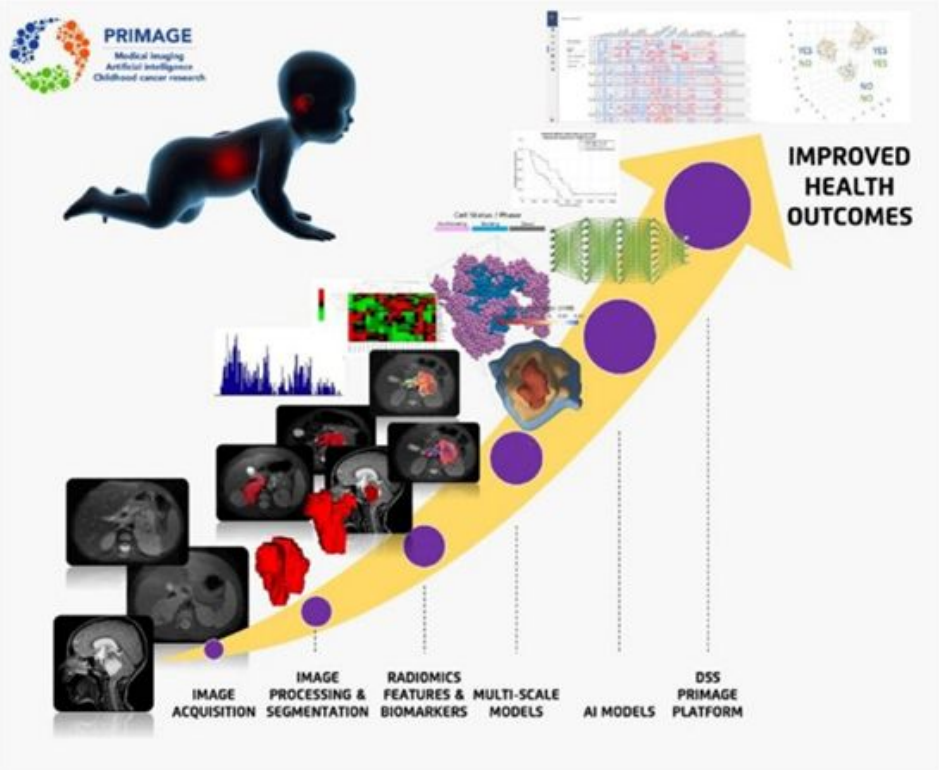
The University Of Sheffield.

PRIMAGE
Medical imaging
Artificial intelligence
Childhood cancer research

Horizon 2020
European Union Funding
for Research & Innovation

Objectives

1. PRIMAGE project.
2. Neuroblastoma.
3. First multicellular model of neuroblastoma.
4. Calibration.
5. Clonal competition.
6. Targeted therapies.



Martí-Bonmatí, Luis, et al. "PRIMAGE project: predictive in silico multiscale analytics to support childhood cancer personalised evaluation empowered by imaging biomarkers." *European radiology experimental* 4.1 (2020): 1-11.

Decision support system for the clinical management of malignant solid tumours.

PRIMAGE project: predictive *in silico* multiscale analytics to support childhood cancer personalised evaluation empowered by imaging biomarkers

Luis Martí-Bonmatí[1*], Ángel Alberich-Bayarri[2], Ruth Ladenstein[3], Ignacio Blanquer[4], J. Damian Segrelles[4], Leonor Cerdá-Alberich[5], Polyxeni Gkontra[5], Barbara Hero[6], J. M. García-Aznar[7,8], Daniel Keim[9], Wolfgang Jentner[9], Karine Seymour[10], Ana Jiménez-Pastor[2], Ismael González-Valverde[2], Blanca Martínez de las Heras[11], Samira Essiaf[12], Dawn Walker[13], Michel Rochette[14], Marian Bubak[15], Jordi Mestres[16], Marco Viceconti[17], Gracia Martí-Besa[5], Adela Cañete[11], Paul Richmond[13], Kenneth Y. Wertheim[13], Tomasz Gubala[15], Marek Kasztelnik[15], Jan Meizner[15], Piotr Nowakowski[15], Salvador Gilpérez[18], Amelia Suárez[18], Mario Aznar[18], Giuliana Restante[19] and Emanuele Neri[19]

I contributed the first multicellular model of neuroblastoma to the project.

This talk is about what I did with the model <u>outside PRIMAGE.</u>

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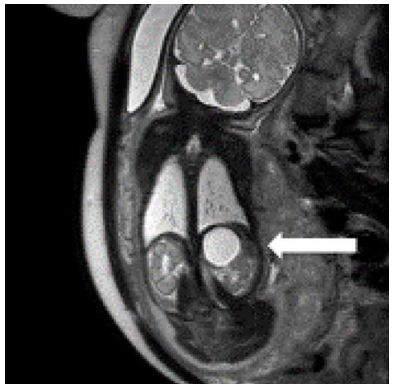
Neuroblastoma

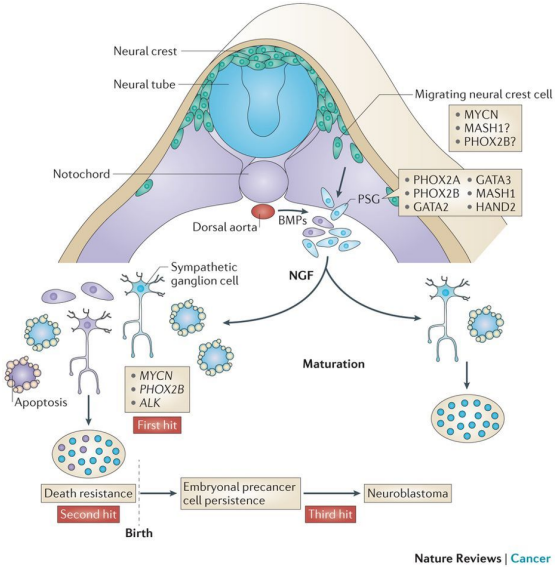


Louis, Chrystal U., and Jason M. Shohet. "Neuroblastoma: molecular pathogenesis and therapy." *Annual review of medicine* 66 (2015): 49.

1. Adrenal medulla is the usual primary site.

2. Most common extracranial solid tumour in children.

3. 15 % of cancer-related deaths in this population.



Marshall, Glenn M., et al. "The prenatal origins of cancer." *Nature Reviews Cancer* 14.4 (2014): 277-289.

1. Neural crest, transient in the embryo.

2. Differentiate into different cell types.

3. Sympathetic nervous system.

4. MYCN amplification and ALK activation turn them into neuroblastoma cells.

INRG Stage	Age (months)	Histologic Category	Grade of Tumor Differentiation	*MYCN*	11q Aberration	Ploidy	Pretreatment Risk Group
L1/L2		GN maturing; GNB intermixed					A Very low
L1		Any, except GN maturing or GNB intermixed		NA			B Very low
				Amp			K High
L2	< 18	Any, except GN maturing or GNB intermixed		NA	No		D Low
					Yes		G Intermediate
	≥ 18	GNB nodular; neuroblastoma	Differentiating	NA	No		E Low
					Yes		H Intermediate
			Poorly differentiated or undifferentiated	NA			
				Amp			N High
M	< 18			NA		Hyperdiploid	F Low
	< 12			NA		Diploid	I Intermediate
	12 to < 18			NA		Diploid	J Intermediate
	< 18			Amp			O High
	≥ 18						P High
MS	< 18			NA	No		C Very low
					Yes		Q High
				Amp			R High

Sokol, Elizabeth, and Ami V. Desai. "The evolution of risk classification for neuroblastoma." *Children* 6.2 (2019): 27.

1. Low risk, spontaneous regression.

2. High risk, 50 % relapse.

3. MYCN amplification is a bad sign.

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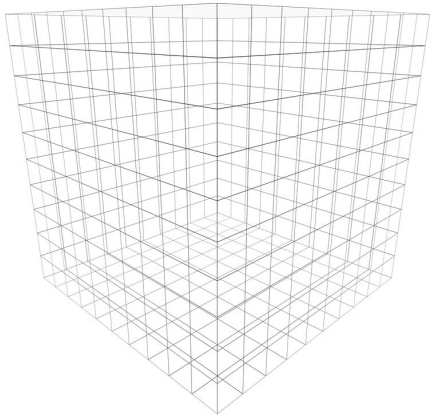
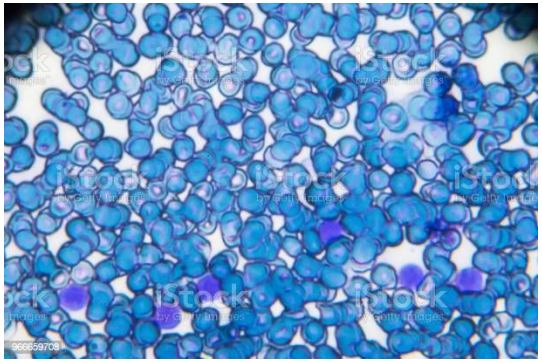
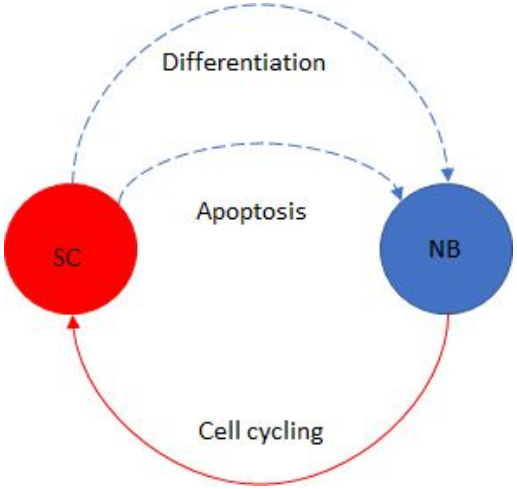
Multicellular model







Continuous automaton to voxelate the microenvironment.

1. Spatial distributions of cells and extracellular matrix.
2. Concentration dynamics of drugs and nutrients (uniform).

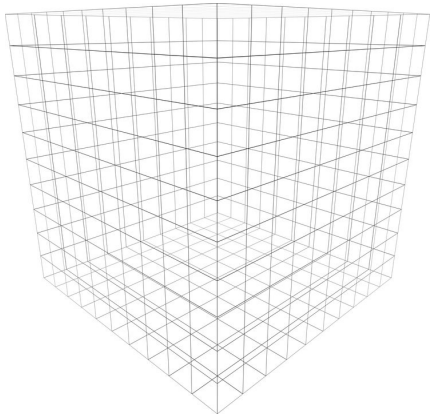
Discrete agents.

1. Neuroblasts and Schwann cells.
2. Cell cycling and death.

Agent attributes.

1. Mutations.
2. DNA status.
3. Gene expression levels.

Multicellular model



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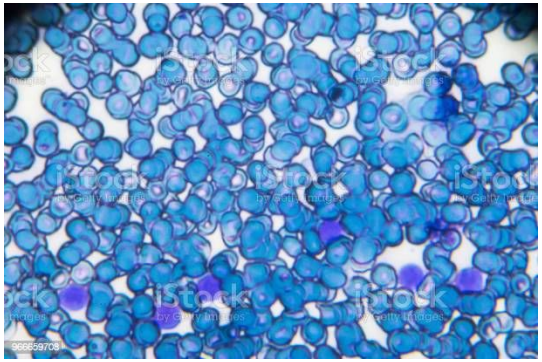

Discrete agents.

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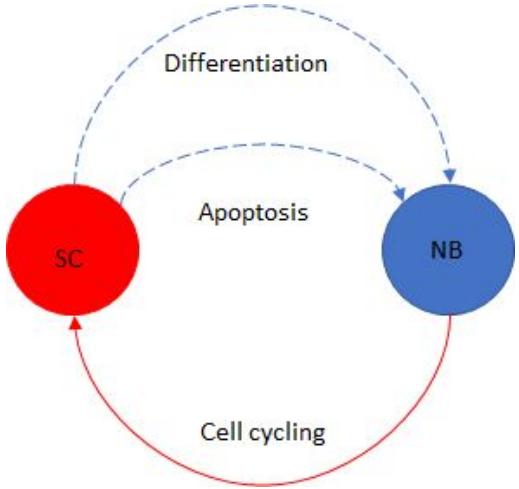
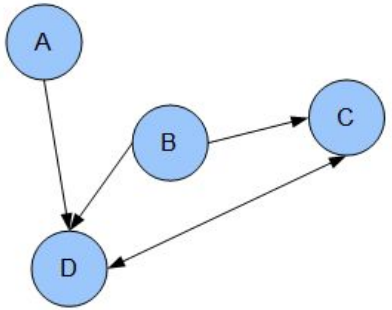
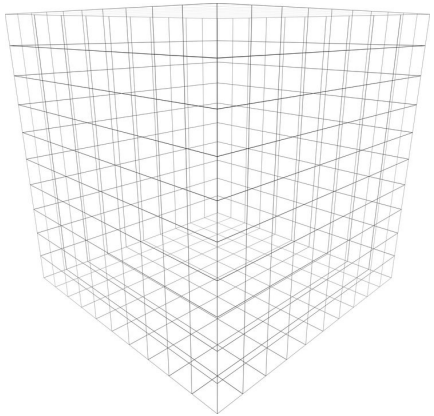




20 gene products.Telomerase, ALT, MYCN, MAPK/RAS pathway, JAB1, CHK1, CDS1, CDC25C, ID2, IAP2, HIF, BNIP3, VEGF, p53, p73, p21, p27, Bcl-2/Bcl-xL, BAK/BAX, and CAS.

Multicellular model



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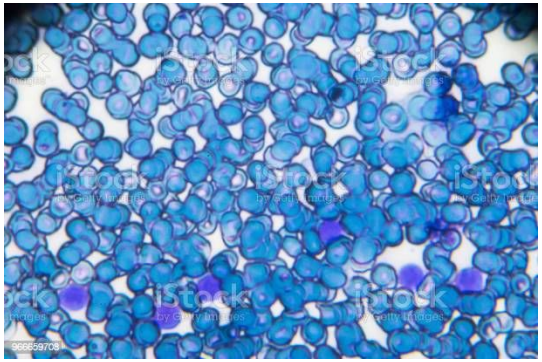

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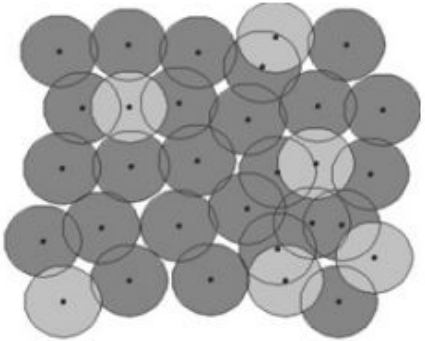

Centre-based mechanical model.

1. Resolve agent-agent overlap and contact inhibition.

2. Linear force law.

3. Equation of motion.

Stochastic simulation algorithm

1. Each agent senses the microenvironment and its neighbouring agents, modifies its behaviour, and updates its attributes.
2. Resolve agent-agent overlap using the mechanical model.
3. Modify the microenvironment by considering the agents collectively.
4. Back to step 1.

A series of Bernoulli trials. For example, is the MAPK/RAS pathway active?

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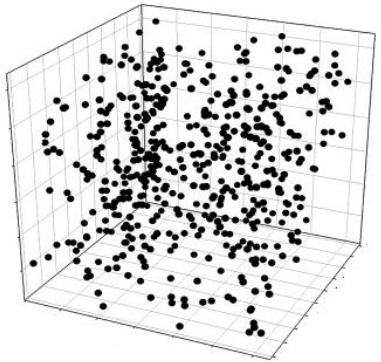
Calibration

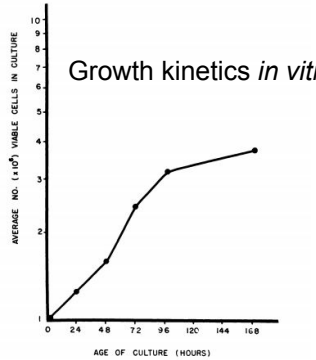


Latin hypercube sampling.

1. 3000 combinations of 20 fitting parameters.

2. Minimised differences between simulation results and *in vitro* data.

3. Refined calibrated parameters for *in vivo* use.



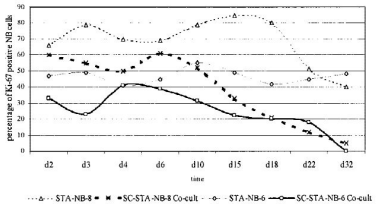
Growth kinetics *in vitro*.

Tumilowicz, Joseph J., et al. "Definition of a continuous human cell line derived from neuroblastoma." *Cancer research* 30.8 (1970): 2110-2118.

	Three-stage fit	95% CI	Direct fit	95% CI
Maximum oxygen consumption rate, q_{max} (mmHg · s^{-1})	17.5	15.3–25.1	16.3	15.3–17.9
P_{O_2} for 50% drop in consumption, $P_{50,q}$ (mmHg)	2.7	0.0–12.5	1.6	1.2–2.1
Maximum misonidazole binding rate, $k_{b,0}$ ($\times 10^{-4}$ s^{-1})	4.5	3.9–4.9	4.4	2.5–5.3
P_{O_2} for 50% drop in binding, $P_{50,b}$ (mmHg)	1.4	0.3–2.6	1.4	1.1–2.5
P_{O_2} for 50% necrosis, $P_{50,n}$ (mmHg)	1.2	0.1–4.9	1.0	0.4–1.2

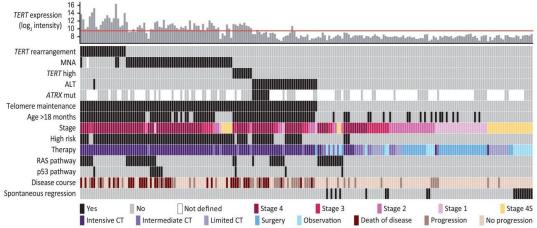
Warren, Daniel R., and Mike Partridge. "The role of necrosis, acute hypoxia and chronic hypoxia in 18F-FMISO PET image contrast: a computational modelling study." *Physics in Medicine & Biology* 61.24 (2016): 8596.

Extent of necrosis during hypoxia *in vitro*.



Ambros, Ingeborg M., et al. "Neuroblastoma cells provoke Schwann cell proliferation in vitro." *Medical and Pediatric Oncology: The Official Journal of SIOP—International Society of Pediatric Oncology (Societé Internationale d'Oncologie Pédiatrique)* 36.1 (2001): 163-168.

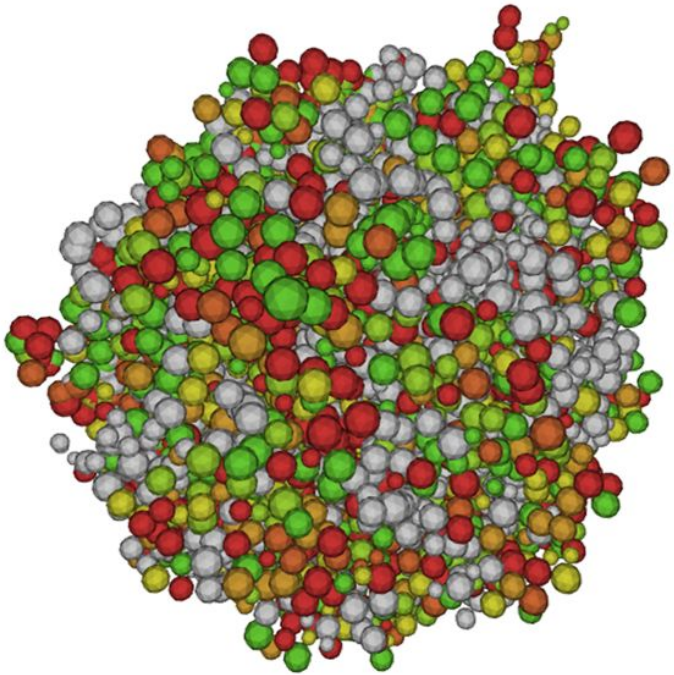
Interactions between neuroblastic and Schwann cells *in vitro*.



Ackermann, Sandra, et al. "A mechanistic classification of clinical phenotypes in neuroblastoma." *Science* 362.6419 (2018): 1165-1170.

Clinical outcomes associated with different mutations.

Calibration



Costly simulations.
1. Millions of agents.
2. Four months in a patient's life.
3. Stochastic simulations.

Simulations on GPUs.

1. FLAMEGPU and FLAMEGPU2 were used to generate optimised CUDA code.

2. 3000 time steps took up to 10 minutes.

3. Calibration took 40 days in total.

Hardware: 2 TITAN V GPUs, 1 TITAN XP GPU, and 1 TITAN RTX GPU.

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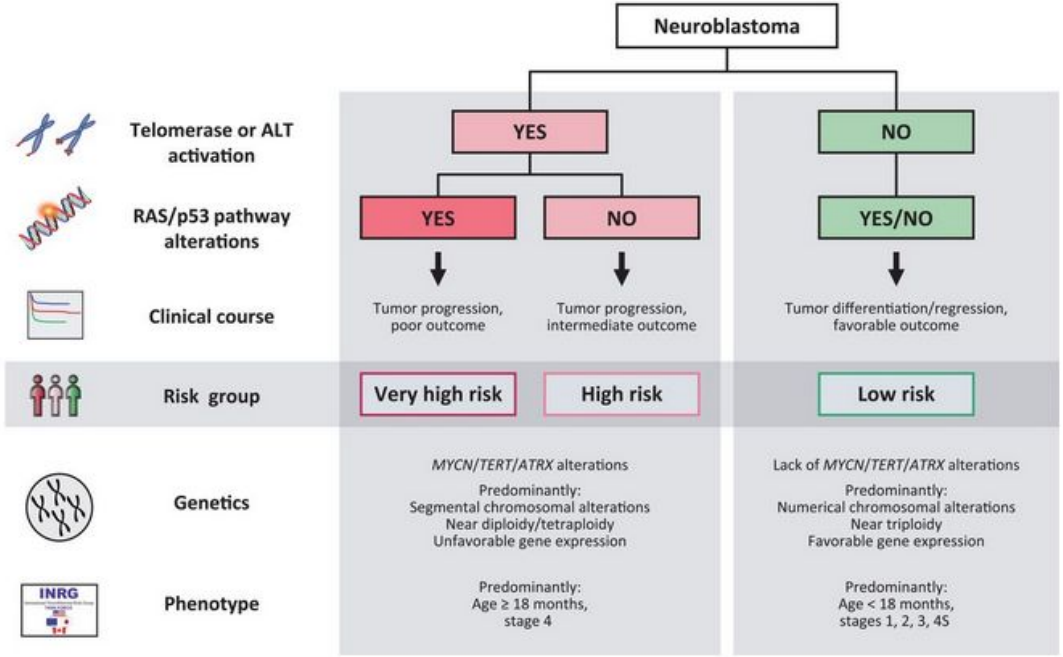
Clonal competition

Clonal composition.
1. Four clones.
2. Each clone has six subclones.

Clones: MYCN amplification, TERT rearrangement, ATRX inactivation, and wild type.

Subclones: combinations of p53 inactivation and ALK activation.

Macroscopic features.
1. Oxygen level.
2. Abundance of Schwann cells.

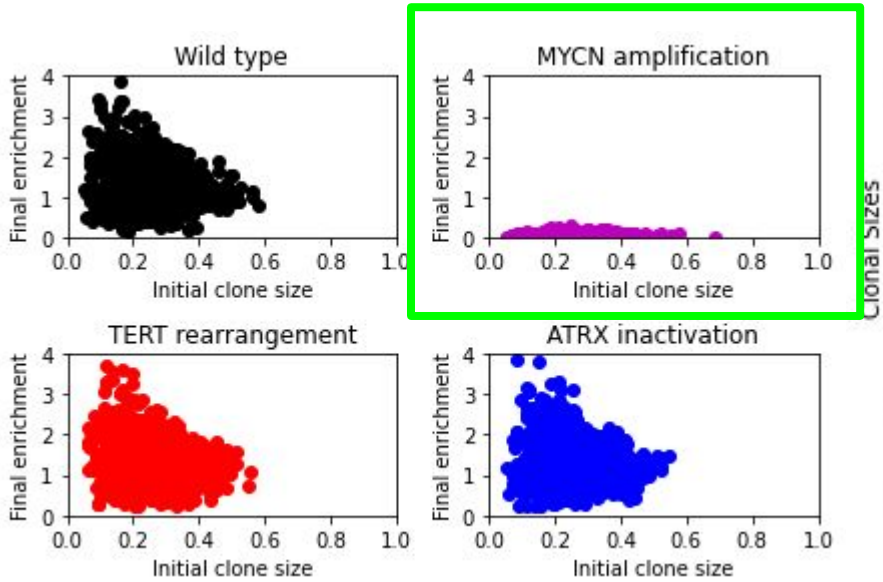


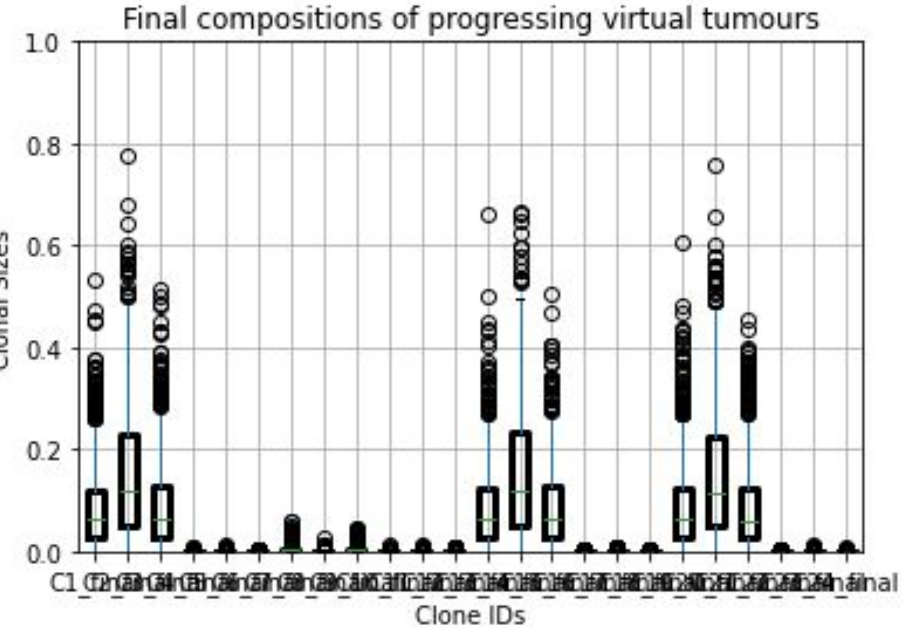
Ackermann, Sandra, et al. "A mechanistic classification of clinical phenotypes in neuroblastoma." *Science* 362.6419 (2018): 1165-1170.

Created 1200 virtual tumours with arbitrary clonal compositions and macroscopic features.

1155 progressing cases.

Clonal competition





MYCN-amplified clone died!

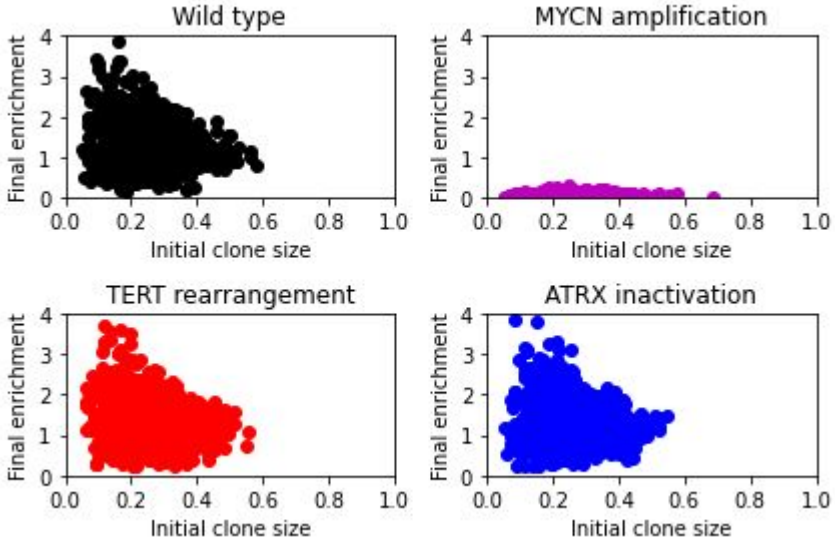
The other three expanded similarly.

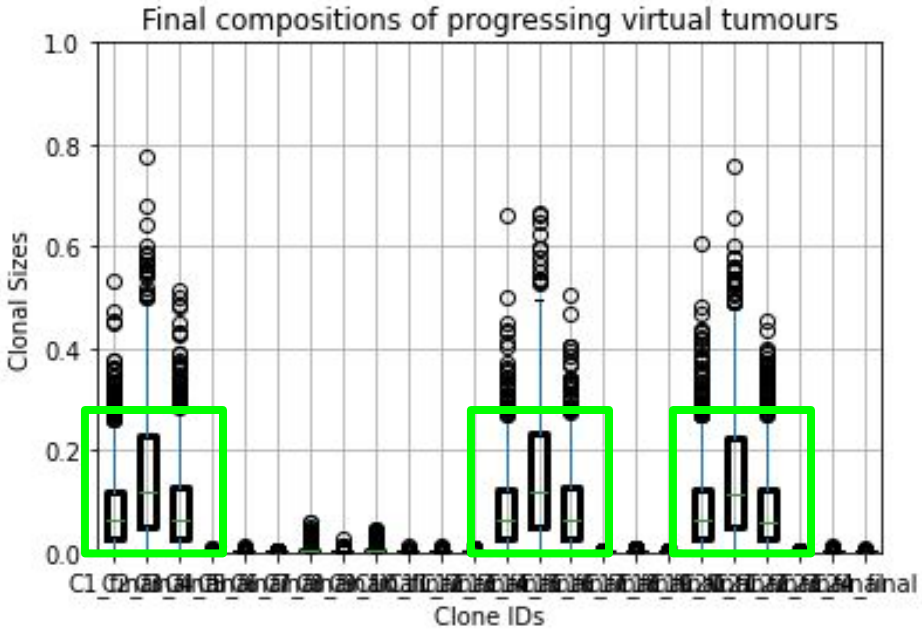
MA versus WT: p-value < 0.1 %
1. Student's t-test.
2. Permutation test.

ANOVA: p-value > 25 %
1. F-test.
2. Permutation test.

1155 progressing cases.

Clonal competition





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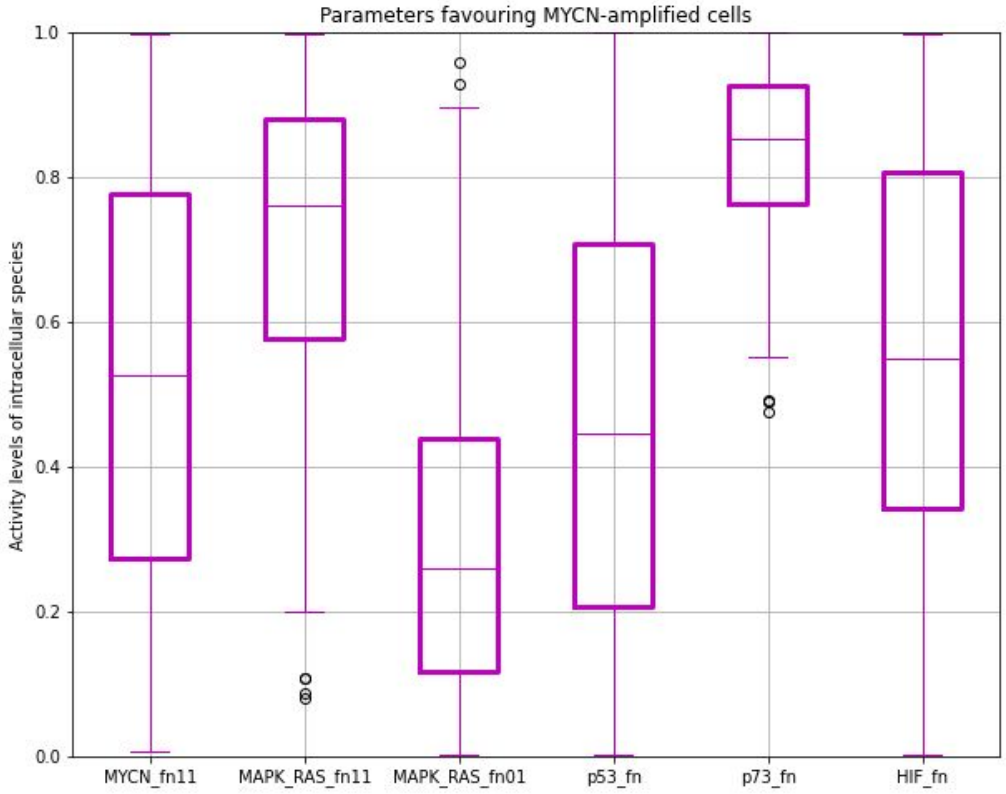
The nine growing subclones all had their p53 intact!

Clonal competition

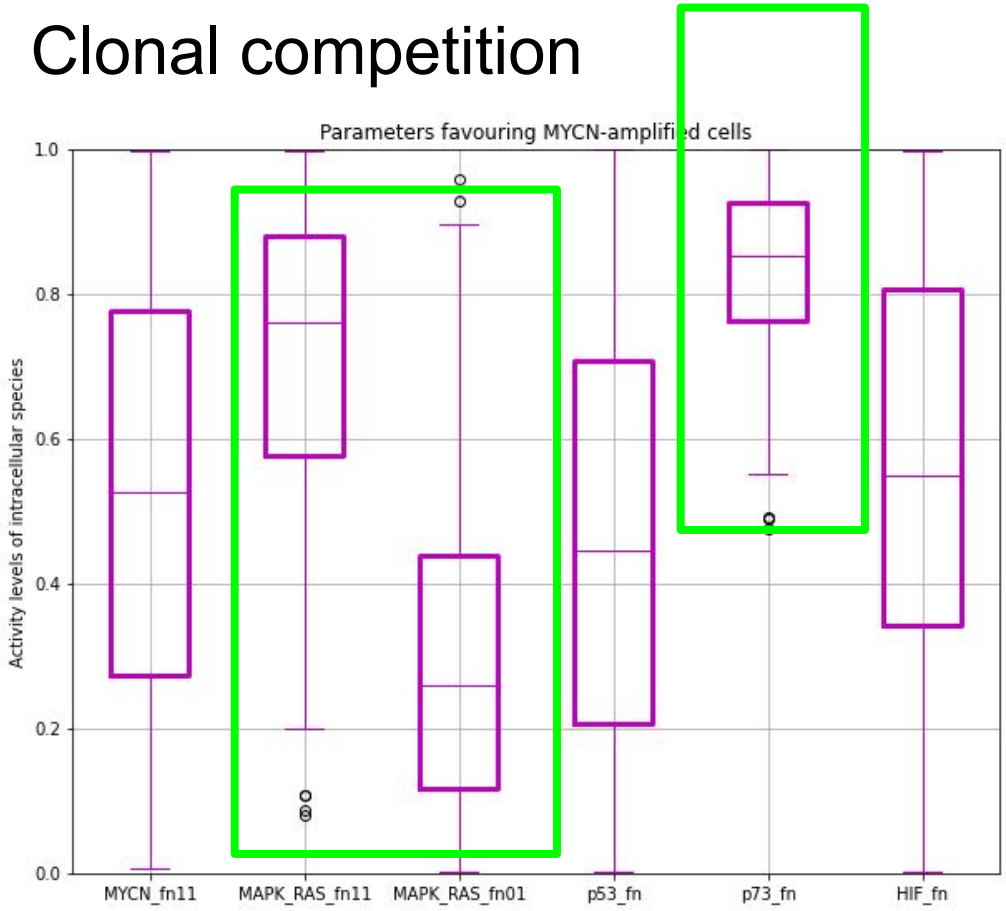


One virtual tumour. Fixed its clonal composition and macroscopic features.

Sensitivity analysis. 1000 combinations of gene expression levels, including MYCN, MAPK/RAS, p53, p73, and HIF.

283 cases where the MYCN-amplified clone expanded drastically.

Clonal competition



283 cases where the MYCN-amplified clone expanded drastically.

1. **p73**, which belongs to the same family, must be active enough to **compensate for the inactivated p53**.

2. **MYCN amplification** must boost MAPK/RAS signalling (**cell cycling**) more than **ALK activation**.

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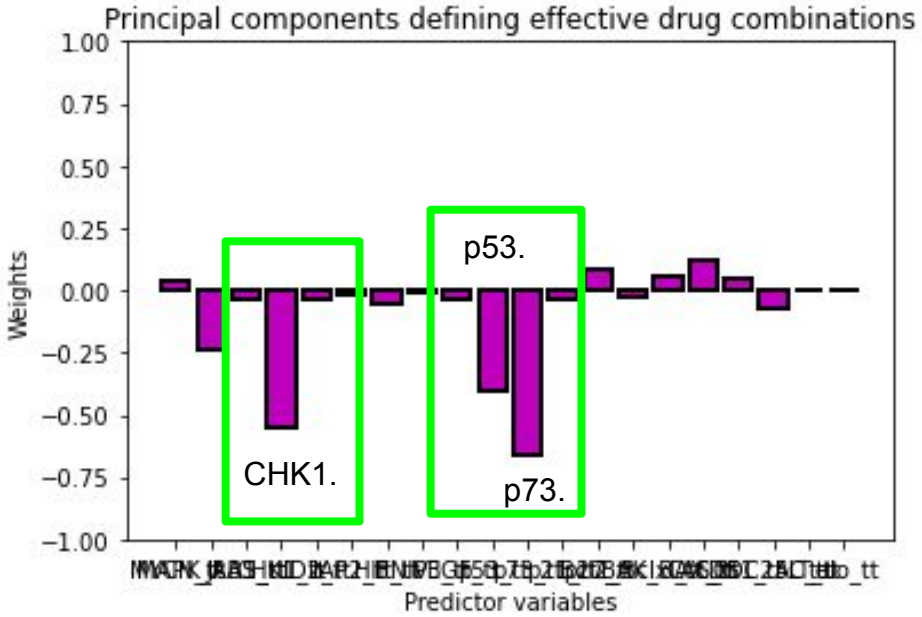
Targeted therapies

Step 1. Created a virtual tumour with one large MYCN-amplified clone only.

Step 2. Chose gene expression levels favouring the MYCN-amplified clone.

Step 3. Tested 1000 combinations of drugs targeting the 20 gene products.



Weights of the first principal component.

Inhibiting CHK1, p53, and p73 shrinks the MYCN-amplified clone.

Consistent with sensitivity analysis, as CHK1 switches on p73.

20 gene products.Telomerase, ALT, MYCN, MAPK/RAS pathway, JAB1, CHK1, CDS1, CDC25C, ID2, IAP2, HIF, BNIP3, VEGF, p53, p73, p21, p27, Bcl-2/Bcl-xL, BAK/BAX, and CAS.

Conclusions

Built, calibrated, and validated the first multicellular model of neuroblastoma.

MYCN-Amplified clone requires p73 and enhanced cell cycling (MAPK/RAS signalling) to thrive.

Drugs targeting CHK1, p53, and p73 are effective against MYCN-amplified clone.